# Piscirickettsia salmonis
# Information sheet

**This information sheet has been prepared from a variety of sources to provide current and unpublished scientific information on Piscirickettsia salmonis relevant to Tasmania.**

**Unfortunately, much of the Australian information available via the internet is outdated. This information sheet focuses on certain topics and does not attempt to describe all aspects of this disease in detail.**

## Summary

In 2024, collaboration between the Centre for Aquatic Animal Health and Vaccines and the Australian Centre for Disease Preparedness (CSIRO, Geelong), facilitated advanced genomic analyses of the bacteria which resulted in the isolation of the East Coast strain of the Tasmanian Rickettsia-Like Organism (TRLO-EC) being reclassified as Piscirickettsia salmonis (P. salmonis). This work was therefore able to determine that P salmonis has in fact been present in Tasmanian east coast waters since at least 2021 and in the south east zone (the 'channel') since 2023 based on samples submitted at those times. This means that this bacterium is naturally occurring in the east and southeast coastal waters and it is no longer regarded as an exotic emergency disease in Tasmania.

## The pathogen

P. salmonis is a facultative intracellular, bacterial, epizootic pathogen of salmonids, first identified in Chile in 1989 and later identified in Canada and several European countries (Fryer and Mauel, 1997). This Gram-negative, pleomorphic, coccoid bacterium replicates within membrane-bound cytoplasmic vacuoles in the cells of infected fish (Fryer et al., 1990).

Piscirickettsiosis, the disease caused by P. salmonis, is also known as salmon rickettsia syndrome and salmonid rickettsial septicaemia (SRS). There are several type strains –including LF-89 (the most virulent Chilean strain) and EM-90. The Tasmanian P. salmonis is most similar to members of the EM-90 genogroup and is closely related to the Tasmanian Rickettsia-Like Organisms.

P. salmonis disease has subsequently been observed in other salmonid species in saltwater (Chinook salmon, Atlantic salmon, rainbow trout and masu salmon) across many parts of the world. It is now known that rickettsia-like organisms affect marine fish over broad host and geographic ranges, acknowledging that many of these species are capable of, or have, marine and freshwater life cycles.

The complete genomic sequence of P. salmonis Chile isolate (LF-89T strain) was obtained in 2001 (Valenzuela et al., 2001). LF-89 has also been found in several non-salmonid hosts such as the white sea bass (Atractoscion nobilis) and other wild species that are not present in Tasmanian waters.

The significance of these detections is not known in terms of how the organism survives in the marine environment or the disease impacts on these other non-salmonid species, but P. salmonis is not known at present to be a risk to native Tasmanian marine species.

## Tasmanian Rickettsia Like Organisms

Since 2001, farmed salmonid fish in Tasmania have been susceptible to infection by Tasmanian Rickettsia-like organisms (TRLOs; Corbeil et al., 2005). TRLOs were considered to be composed of three serovars, with variants linked to one of three geographic regions of Tasmania, the southeast, Macquarie Harbour or Tamar River (Morrison et al., 2016). These three closely related TRLOs are also closely related to P. salmonis. TRLO-SE, TRLO-MH and TRLO-TR are now known to form a distinct phylogenomic clade, which most likely represents a novel species of Piscirickettsia. It is anticipated that this species will be named when these data are published in the peer-reviewed literature.

## Phylogeny

Molecular phylogenetic analysis based on sequencing of the 16S rRNA gene categorised P. salmonis in a new family of Piscirickettsiaceae and Genus Piscirickettsia within that family and within the class of Gamma (γ)-proteobacteria. Although they belong to different classes, Piscirickettsia (Gamma-proteobacteria) is morphologically similar to true Rickettsia bacteria (Alpha-proteobacteria). Complete genomic sequences from more strains of P. salmonis as well as the TRLOs, will allow the refinement of their phylogeny. It is now further proposed for these TRLO isolates to join the Genus Piscirickettsia in the Gamma-proteobacteria.

## Ecology – epidemiology

Although it was initially described as obligately intracellular, more recent research has established that P. salmonis can survive as both a free-living bacterium in the marine environment and in laboratory settings. In seawater, free-living P. salmonis can survive for at least 21 days under the right environmental conditions and is capable of forming viable and mucus-tolerant biofilms on non-living surfaces. Survival is highest at around 5 °C and decreases as temperature increases; almost no survival is observed above 25 °C, but this is also an unfavourable temperature for Atlantic salmon.

Outbreaks usually occur after fish are introduced to saltwater at water temperatures between 12°C and 18°C. This temperature range corresponds to that experienced in salmon leases in Tasmania but the disease here to date is primarily seen in mature fish towards the end of their growth cycle in the marine environment and associated with peak summer water temperatures.

P. salmonis does not appear to be able to survive without a host in freshwater environments as it is readily lysed in freshwater. As such it is unlikely to be a threat to Tasmanian freshwater trout and salmon species. Biosecurity standards are in place to prevent the movement of salmonids from marine to freshwater environments. The impact of P. salmonis on the estuarine population of brown trout in Tasmania is difficult to predict but it is expected to be much less than for farmed salmon.

## Human Health

As a marine bacterium P. salmonis is a fish pathogen and does not cause disease in people.

The optimum temperature range means that it does not pose a threat to warm-blooded species including humans. It's been shown that there is sub-optimal growth of P. salmonis at 29°C (Henriquez et al., 2013) and no growth at 30°C (Mikalsen et al., 2008). Generally advice remains to not handle or consume the dead fish material; avoid activities that will bring you into contact with the material , and wash your hands with soap and water if you do.

With the specific growth requirements for a marine bacterium, P. salmonis is a fish pathogen and does not cause human or terrestrial animal disease, or any food safety risk.

## Diagnosis

Diagnosis of piscirickettsiosis is based on a range of procedures. Presumptive diagnosis is made following clinical and pathological observations. P. salmonis is confirmed following histopathological examination, isolation (growth) in tissue culture on solid medium or in liquid medium, combined with identification by dot blot serotyping. Identification of bacteria by mass spectrometric (matrix-assisted laser desorption ionization – time-of-flight; MALDI-TOF) analysis is a key part of the initial clinical diagnostic laboratory processes in Tasmania and across the globe.

In addition, a polymerase chain reaction (PCR) technique for the rapid identification of the Australian isolate of P. salmonis is being developed. Whole genome sequencing (WGS) and analysis of cultured organisms is currently required to confirm the exact strain of bacteria.

## Tasmanian timeline

Since 2001, farmed salmonid fish in Tasmania have been susceptible to infection by Tasmanian Rickettsia-like organisms (TRLOs; Corbeil et al., 2005). The initial diagnosis of P. salmonis came from whole genome sequencing (WGS) in early 2024. WGS of the Tasmanian collection of 41 TRLO isolates held at the Animal Health Laboratory (AHL), Mount Pleasant, was initially undertaken to resolve phylogenetic relationships with closely related taxa and develop a TRLO specific molecular diagnostic assay (PCR).

The project began by completing a pilot assessment of in-house Oxford Nanopore Technology (ONT) sequencing methods using three so-called TRLO-EC isolates that were isolated in pure culture from samples were delivered to the AHL in February 2024, from Atlantic salmon grown at a lease in southern Tasmania. They were submitted under suspicion that fish were affected by Tasmanian Salmonid Rickettsiosis (TSR), the disease associated with Tasmanian RLOs. Molecular testing showed that 4/4 samples were RLO/P. salmonis qPCR (Corbeil et al., 2003) positive. Bacteria were isolated in pure culture from three liver samples and their identity confirmed as 'TRLO-EC' by RLO/P. salmonis qPCR (Corbeil et al., 2003) and dot blot serotyping.

From this WGS analysis, the P. salmonis organism has been determined retrospectively in 2024 to have been in Tasmanian east coast waters since at least 2021 indicating that the pathogen may have been circulating for five or more years. It is possible that it was in east coast waters from 2019.

## Pathology

A range of gross signs of disease may be present in salmonids infected with P. salmonis in common with many systemic, chronic inflammatory diseases of salmonids. Liver pathology (abscesses) may be the only definitive sign seen macroscopically. Confirmation requires testing in a fish health laboratory such as the AHL, Mount Pleasant.

It is worth noting that isolation of P. salmonis in culture has occurred within the AHL, a physical containment level 2 (PC2) facility. According to AS/NZS 2243.32, culture of risk group 3 pathogens, including P. salmonis, is restricted to PC3 laboratories. However, at the time of isolation, cultures were obtained on the understanding that they were a variant TRLO (i.e. risk group 2) and not an exotic risk group 3 pathogen. It is anticipated that there will be reclassification of the AS/NZS 2243.3 risk group to which P. salmonis is assigned.

## Antibiotics

A bacterial outbreak is managed by industry veterinarians through the approved use of antibiotics (primarily Oxytetracycline to date), but only rarely and when necessary – no different to any other animal illness.  Veterinarians are required to report the use of antibiotics in the marine environment in Tasmania to the Tasmanian Chief Veterinary Officer and the Director of the Environment Protection Authority. If fish were successfully treated with antibiotics they would have to be held for a certain calculated period (approximately two months) before they can be harvested for human consumption. This period allows the depletion of antibiotic residues to levels below Australian food safety standards levels, and they would be perfectly healthy to eat.

## Vaccine

A vaccine has been developed by the Centre for Aquatic Animal Health and Vaccines, Mount Pleasant, and is used as a key strategy to protect salmon from this bacterium. The vaccine protects Atlantic salmon against eight bacteria, including P. salmonis (TRLO-EC) and is expected to assist the Tasmanian salmon industry in mitigating against the immediate threat posed by P. salmonis. However, no vaccine can be regarded as being completely protective and there are a number of factors that may degrade vaccine immunity. Scientific studies continue into the characteristics of the bacterial isolates and their relevance for fish vaccines. Fish can only be vaccinated by injection when they are still quite small in the freshwater hatchery.

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